INTERNATIONAL FORMULA COUNCIL

1100 Johnson Ferry Road, Suite 300 ▪ Atlanta, GA 30342
(404) 252-3663 ▪ Fax (404) 252-0774 ▪ E-mail: info@infantformula.org ▪ www.infantformula.org

May 29, 2008

Dr. Barbara Shane
NTP Office of Liaison, Policy and Review
NIEHS
111 T.W. Alexander Drive
Room A322
Research Triangle Park, NC 27709

Re: International Formula Council's Comments Regarding the NTP-Brief on Bisphenol A

Dear Dr. Shane:

The International Formula Council* (IFC) is responding to the notice published in the April 15, 2008 *Federal Register* by the National Toxicology Program (NTP) requesting comments concerning the NTP Draft Brief, which was released on April 14, 2008. The purpose of this letter is to address several concerns regarding the NTP Draft Brief on bisphenol A (BPA). These concerns are as follows:

1. The inclusion of studies in which a subcutaneous route of BPA administration was used

A key difference between the NTP Center for the Evaluation of Risks to Human Reproduction (NTP-CERHR) Expert Panel Report and the NTP Draft Brief on BPA is that, in the latter document, studies with a subcutaneous route of administration were considered. The rationale for the inclusion of such studies is not clear given that human exposure to BPA is overwhelmingly oral. The subcutaneous administration of BPA ignores the significant degree of glucuronidation of BPA that occurs either by intestinal cells or hepatic first pass metabolism. Because glucuronidation eliminates the potential estrogenic activity of BPA, this consideration is extremely important.

A possible explanation for this sudden change of philosophy is the results of a study by Taylor et al. (2008). In this study, two doses of ^{3}H-BPA (35 and 395 mcg/kg bw/day) were administered to newborn mice (postnatal day 3) by both the oral and subcutaneous routes. Serum levels of ^{3}H-BPA were monitored for 24 hours after administration of the dose. The authors noted no

* The International Formula Council is an association of manufacturers and marketers of formulated nutrition products, e.g., infant formulas and adult nutritionals, whose members are predominantly based in North America. IFC members are: Abbott Nutrition; Mead Johnson Nutritionals; Nestlé Nutrition – USA; and Wyeth Nutrition.

difference in serum ^{3}H-BPA levels between the two routes of administration and concluded "this finding sets aside the belief that non-oral administration of BPA renders data as not suitable for consideration of the hazard posed by low dose BPA exposure during neonatal life" (page 1, abstract). This sweeping conclusion, based on the results of just one animal study, is not justified for several reasons.

The first problem with this study is that there was no actual determination of free versus conjugated BPA. The statement was made that BPA extraction from plasma using tert-butyl methyl ether would extract free BPA (non-glucuronidated, but bound to serum proteins) but not water-soluble conjugates (such as glucuronidated BPA). By definition, then, this method excludes any potential conjugates of BPA, such as the glucuronidated or sulfated form. This is a serious limitation because the authors hypothesize that newborns do not have the ability to conjugate BPA. Not measuring the conjugates makes it impossible to evaluate this hypothesis. It is well known that neonates possess a lower capability to conjugate BPA than do adults. However, even the "low" dose of BPA used in this study (35 mcg/kg bw/day) is 35 times higher than the typical infant exposure of 1.0 mcg/kg bw/day estimated by the NTP-CERHR Expert Panel (NTP Draft Brief, page 33). While it is recognized that fetal and neonatal beta-glucuronidase activity may be reduced compared with mature animals, the presence of some residual beta-glucuronidase activity, perhaps enough to deal with the minute amounts of BPA associated with typical dietary exposure, cannot be ruled out. Several human studies have indicated that drugs such as morphine and tramadol can be glucuronidated to some degree by the fetal or neonatal liver (Allegaert et al., 2006; Pacifici et al., 1982). Because Taylor et al. (2008) did not attempt to measure conjugated forms of BPA, the degree of potential BPA glucuronidation versus the complete absence of BPA glucuronidation cannot be evaluated from their study.

A second problem with the Taylor et al. study is that even at the low BPA dose C_{max} (1.78-2.60 ng/mL plasma) and the high BPA dose C_{max} (13.10-14.82 ng/mL plasma) values reported, these values represent <0.5% of the amount of BPA actually administered to the animals (assuming a body weight of 2.4 g and total blood volume of about 0.2 mL). The fate of the remainder of the BPA (metabolism to conjugates, urinary excretion, tissue storage) was not explored. Only plasma BPA levels were measured, which gives virtually no information on the disposition of the BPA dose.

A third potential limitation is that the choice and age of the model animal may not be representative of the human. Although comparing relative ages of mice or rats versus humans is difficult, it is likely that the first week to 10 days of a rat or mouse's life is roughly equivalent to the period of late gestation in humans (Quinn, 2005). Thus, the 3-day old mouse pup used in this study is probably the equivalent of a human fetus in late gestation. This is significant because during the gestational period, the fetus will be largely protected from free BPA via the maternal capability for BPA glucuronidation. It is also well known that rodents and humans excrete conjugated BPA much differently, with the former excreting it mainly via the biliary route while the latter excrete it primarily in the urine (Volkel et al., 2002). It is not certain whether the lack of first-pass hepatic conjugation, as suggested by this study, also applies in humans, and if there is a low conjugation efficiency in neonatal humans, whether the target tissue sensitivity to BPA corresponds to that of the neonatal mouse.

Finally, the choice of the mouse, while familiar to the investigators and relatively inexpensive, is not well justified. It is acknowledged that there are ethical constraints regarding conducting such

invasive BPA studies in humans, necessitating an animal model. However, it is unclear why Taylor et al. (2008) would not use either a primate model or another animal model that is more closely representative of the human gastrointestinal tract (e.g., a piglet) instead of the mouse. It is difficult to imagine that a 2.4 g mouse pup is very representative of a 7-8 pound human infant. In addition, the mouse pups had to be sacrificed to obtain the amount of blood necessary for this study. This prevented the authors from examining BPA clearance via urine or other routes over time within a particular animal. Use of a larger animal would have allowed for more frequent blood sampling within an animal while it is still alive and would also allow for evaluation of urinary BPA clearance. The animal could then be sacrificed later, if needed, to examine tissue levels of BPA.

In summary, subcutaneous injection of BPA is unrepresentative of human exposure. Further, the only study that has attempted to equate subcutaneous and oral BPA administration had numerous and serious limitations that prevent valid conclusions on this issue from being drawn. Thus, there appears to be no reasonable justification for including studies of subcutaneous administration in the NTP Draft Brief and the conclusions of the CERHR Expert Panel are more in accordance with the data that presently exist.

2. Upgrading the level of concern from "minimal" to "some" for BPA effects on the prostate, mammary gland, or early puberty (compared with NTP-CERHR Expert Panel Report)

This upgrade in the concern level is unnecessary based on a review of the literature and is contradictory with several statements made in the NTP Draft Brief. With regard to the advancement of puberty associated with "low" dose BPA, one of the most cited works is that of Howdeshell et al. (1999). This study reported a ~2.5 day acceleration in puberty in the offspring of pregnant CD-1 mice exposed to 2.4 mcg/kg bw/day BPA during days 11-17 of gestation. However, as noted by the NTP Draft Brief, this group used a non-standard method of assessing puberty (e.g., the interval between vaginal opening and first estrus). In addition, the study was only published as a brief research communication in Nature, meaning that the explanation of the methodology was not as comprehensive as it would be in a full research article.

The NTP Draft Brief also reports a study by Ryan et al. (2006) showing a 4.5-day acceleration in puberty in mice, as assessed by the time interval from birth to first estrus. However, it should be noted that the "high" dose of BPA fed during gestation (200 mcg/kg bw/day), which is roughly 200 times typical human exposure, was the only dose to find an effect. The 20-mcg/kg bw/day dose-much, which is more representative of human exposure, showed no effect. Ryan et al. states that there was a main effect of the reduction of errors, but when one examines the plotted data, they suggest there is no difference from the overall pattern observed for control animals, and there was no difference reported on arm choice of first error, or on Barnes maze for either 2 or 200 mcg/kg groups), so the only clear difference related to BPA was a preference for a light chamber in the 200 mcg/kg group.

Another study, cited by the NTP Draft Brief, in mice (Honma et al., 2002) showed a ~1 day advancement in puberty, but this was associated with subcutaneous administration of BPA (20 mcg/kg bw/day)-an irrelevant route of exposure. To bolster the argument against low dose BPA effects on puberty, well designed and adequately powered studies in both rats (Ema et al., 2001) and mice (Tyl, et al., 2008) showed no effects of BPA on the advancement of puberty. The sum

total of all of these "low-dose" studies is that there is no consistent effect of such doses (delivered orally) on the advancement of puberty in either rats or mice.

With regard to effects on the prostate and mammary gland, the findings of increased prostate weight in the offspring of pregnant Cf-1 mice exposed to 2 or 20 mcg/kg bw/day BPA during days 11-17 of gestation (Nagel et al, 1997) can be criticized for small sample sizes (n=7-11 animals per treatment group) and for the numerous failures of other laboratories to replicate these findings in better-designed, more adequately powered studies (Ashby et al., 1999; Cagen et al., 1999; Nagao et al., 2002). A recent government-funded study from Howdeshell et al. (2008) also showed no effects of 2, 20, or 200 mcg BPA/kg bw/day on male reproductive organ weights in the Long Evans Hooded rat. The NTP Draft Brief states: "In brief, the NTP believes that the overall conclusions of the Bisphenol A Subpanel of the NTP Low-Dose Peer Review remain valid with respect to "low" dose effects on prostate weight, i.e., increased prostate weight cannot be considered a general or reproducible finding" (p. 25). A further statement regarding prostate cancer reads as follows: "The evidence is not sufficient to conclude that bisphenol A is a rodent prostate gland carcinogen or that bisphenol A presents a prostate cancer hazard to humans" (p. 23).

With regard to BPA effects on the mammary gland, the NTP Draft Brief did cite some rodent studies in which perinatal exposure to BPA via a "subcutaneous mini-pump at doses of 0.0025 to 1.0 mg/kg bw/day causes tissue changes ("lesions") in the mammary gland that may signal an increased susceptibility to develop mammary gland tumors later in life" (p. 20). However, the poor rationale for including studies of subcutaneous BPA administration has already been discussed at length. In addition, a dose of 1.0 mg/kg bw/day is 1000-fold higher than typical human exposures, which is irrelevant to risk assessment in humans (especially when delivered by subcutaneous injection). In the presence of this and other studies, the NTP Draft Brief still makes the following statement regarding breast cancer: "The evidence is not sufficient to conclude that bisphenol A is a rodent mammary gland carcinogen or that bisphenol A presents a breast cancer hazard to humans" (p. 20).

In summary, based on the NTP's review of the literature and the statements made by the NTP, there appears to be no consistent evidence for an effect of orally fed, "low" doses of BPA (which is the scenario representative of typical human exposure) on the advancement of puberty or development of either the prostate or mammary gland. Thus, the decision to upgrade the NTP-CERHER Expert Panel Report's level of concern from "minimal" to "some" is perplexing.

3. Criticisms of the studies on neurobehavioral outcomes in "low" dose BPA studies that led to the "some concern" rating for this endpoint

The first criticism of the NTP Draft Brief is the inclusion of studies that use subcutaneous mini-pumps or administration of BPA directly into the brain. As previously stated, the methods of administration are not representative of human exposure.

A second concern is that studies of oral doses of BPA on the normal male-female differences in behavior have so many weaknesses in study design that they have little value in the application to humans. The NTP-CERHR Expert Panel Report reviewed this literature and found that many of the commonly cited studies for these effects (Fujimoto et al., 2006; Dessi-Fulgheri et al., 2002; Farabollini et al., 2002; Kawai et al., 2003) were of such poor quality that they were not to be included in the analysis.

Serious limitations of the various studies in the neurobehavioral literature on BPA include at least one of the following per study:

1) Small sample sizes (generally < 15-20 animals per treatment group)
2) No mention of the blinding of behavioral observers to the research hypothesis or treatments received by the animals
3) Subcutaneous administration of BPA or direct BPA administration into the brain
4) Unclear relevance of observed behaviors in rats/mice to humans
5) Lack of clarity regarding the "adversity" of such behavior changes
6) Inappropriate statistical analysis in which litter effects are not properly accounted for
7) Use of so-called "low" doses, e.g., 50-100 mcg/kg bw/day, that are still well above typical human exposures. The highest potential worst-case exposure is 14-15 mcg/kg bw/day in infants and young children (Tables 10-14 of NTP-CERHR Expert Panel Report), due to lower body weights. However, typical exposures in all age groups are probably closer to 1 mcg/kg bw/day or less.
8) Inconsistent data within a study, such as that found in an article by Palanza et al. (2002). In this study, pregnant CD-1 mice were treated orally with 10 mcg/kg bw/day BPA during gestation days 14-18. Then some of the prenatally treated offspring were again treated with BPA during days 14-18 of their gestation (same dose). This created 4 groups (controls, prenatal BPA exposure, adult BPA exposure, and both prenatal and adult exposure). Dams exposed to BPA either prenatally or during adulthood spent less time nursing their pups and more time out of the nest than controls. However, inexplicably, the group receiving both prenatal and adult exposure was no different from controls. Findings such as this raise the distinct possibility of spurious statistical differences that have no biological relevance.

The NTP Draft Brief concluded that there was evidence for "some concern" for neural and behavioral effects in fetuses, infants, and children at current human exposures based on consistent number of "positive studies" and possible behavioral alterations related to sexual dimorphisms in rodents. We call into question whether the data on neural and behavioral response is consistent. The Expert panel cites 9 papers at the point in the report where they draw their conclusion that there is sufficient data to suggest that BPA causes neural and behavioral alternations in rats and mice; only some of these nine studies address neural and behavioral outcomes. An evaluation of these and other studies examined by the NTP and/or the CERHR Panels follows below.

Nishizawa et al. (2003) reported single-dose BPA-related changes in mRNA levels in some mouse embryos brain regions and reproductive organs at some time points in gestation (but not at others). There was no positive control in this study. Funabashi et al. (2004) reported histological observations on two brain regions stained imunohistochemically for corticotropin releasing neurons. There were sex-related differences in control animals in the stria terminalis and preoptic area. BPA exposure had no effect on the sex difference in the preoptic area, but abolished the sex difference stria terminalis.

Nishizawa et al. (2005) reported a multiple dose study in mice exposed in utero to BPA or estrogen. Levels of a few types of mRNA in a few tissues were shown to be changed. The dose response curve shape was quite variable as well; some results had a 'U" shape (eg. AhR mRNA), some an inverted "U" shape (eg. AhRR in testes), some inversely linear (eg. Arnt mRNA in cerebra, ovaries), and some with no apparent relation to dose. The CERHR Expert Panel

determined that these studies by Nishizawa were adequate, but of limited utility for evaluation. Nagao et al. (1999) reported no change in the area of preoptic reproductive behavior after postnatal BPA. Furthermore, Kwon et al. (2000), reported organ weights after prenatal exposure to BPA and also measured the SDN-POA volume of the brain of female rats, but found no effect of BPA on these measures.

Rubin et al. (2006), exposed mice to subcutaneous administration of 25 or 250 mcg BPA/kg bw/day from day 8 of gestation through day 16 of lactation and measured behavioral changes in an open field test in the offspring at 6-9 weeks of age. This study showed some narrowing of the normal gap between males and females in rears at the wall/5 min, time in center, and time in which the animal is stopped. However, none of the alterations in behavior between males and females had a statistically significant association with BPA

Four studies cited by the CERHR Expert Panel were also specifically cited by the NTP Brief (Ceccarelli et al., Negishi et al., Ryan, and Laviola et al. et al); these studies examined pubertal exposure effects in sexually dimorphic regions of the hypothalamus. These studies noted few effects and are not particularly relevant to the conclusion of BPA posing "some concern" for infants. Among these studies, behaviors were measured differently in every study, including the BPA-related differences in avoidance of electric shock, preference of a light chamber, maze running, and conditioned place preference. Negishi et al., for example, reported that male pups exposed to 100 mcg/kg from GD3-PND 20 had normal behavior. BPA exposed males had a reduced avoidance of electric shock, and an attenuated response to monoamine oxidase inhibitor-induced locomotion. Laviola et al. found no difference in normal behavior among in utero-exposed males, nor were BPA exposed males different than controls in their response to amphetamine-induced behavior, but amphetamine-induced behavior was attenuated among females exposed to BPA in utero, loosing statistically-significant difference observed among controls.

There are 2 additional studies on neural and behavioral outcomes cited by NTP that were not cited by the CERHR Expert Panel. Palanza et al. gave 10 mcg/kg to pregnant rats at gestational day 14-18, or to adults or at both times, and measured the time the females spent later, nursing their own offspring. Compared to unexposed animals, the exposed fetuses spent less time nursing, and the exposed adults time spent less time nursing, but animals exposed in utero and as adults were not different than controls. Gioiosa et al. found some exploratory behavior differences that were all the product of gender-BPA interaction. For males there was no effect of BPA on 4 of 6 measures of behavior; BPA exposed males had a shorted latency before entering a novel compartment and less rearing was observed in the novel compartment. Similarly for females there was no effect of BPA on 4 of 6 measures; BPA exposed females spent a shorted time in the novel compartment and spent more time self grooming. BPA had no effect on either gender for risk assessment or locomotor activity in the novel compartment.

The Palanza study is internally contradictory, as previously mentioned. The addition of the data by Gioiosa et al. allows comparisons of outcome behaviors to other studies. Gioiosa's BPA exposed males had two (of six measured) differences in normal behavior; Negishi's BPA exposed males showed normal behavior. Gioiosa et al. reported that BPA abolished the sex differences to a stressed maze test: males spent more time in the center but not less time or frequency of entry into open arms of a maze, whereas Negishi reported that BPA treatment of males caused an increased residual anxiety to a previous electroshock.

Further, the data on BPA-associated behavior change can be contradicted by well-designed studies. Ema et al. (2001) showed no effects of 0.2, 2, 20, or 200 mcg BPA/kg bw/day on behavior in a two-generation study of Sprague-Dawley rats. The BPA was given via gastric intubation at the onset of a 10- and 2-weeks premating period in F0 males and females, respectively, and continued throughout the mating, gestation, and lactation periods, for two generations. Open field tests, which evaluated ambulation, rearing, grooming, and occurrence or urination and defecation, were conducted on three successive days in all F1 rats at 5-6 weeks of age. There were no BPA-related effects on the normal male-female differences in behavior for either the open field test or for a water-filled multiple T-maze (all F1 offspring measured at 6-7 weeks of age). In addition, there were no effects of BPA on several reflexes (surface righting, negative geotaxis reflex, mid-air righting reflex).

In summary, the existing studies of neurobehavioral effects of BPA suffer from a number of serious weaknesses in design; the weight of evidence does not support a consistent effect and certainly not an adverse one. The data from a well-designed two-generation study in rats, one that received very little criticism from the NTP-CERHR Expert Panel, showed no effects of BPA on the normal behavioral differences between males and females. Finally, there has been no credible evidence from human studies of any behavioral effects in people exposed to BPA either via diet or industrial settings (e.g., spraying of BPA-containing epoxy resins). Given the all of this information against the "low-dose" hypothesis for BPA effects on neurobehavioral endpoints, it is difficult to justify a rating of even "some concern."

4. **Failure of the either the CERHR Expert Panel Report or the NTP Draft Brief to adequately address the weakness of the endocrine disruptor hypothesis in general**

There are several points that merit consideration. The first is that there is absolutely no evidence that "endocrine disruptors" cause harm to humans, especially in the amounts in which they may be present in the environment. However, advocates of the endocrine disruptor hypothesis often make claims about endocrine disrupters causing health problems that, in fact, may not even exist. For example, Carlsen et al. (1992) and Sharpe et al. (1993) argued that there has been a decline in semen quality over the past 50 years and that the exposure to environmental estrogens may be at least partially responsible. The conclusion regarding the decline in semen quality was based on a controversial meta-analysis of 61 sperm count studies. However, a number of subsequent studies have cast considerable doubt over whether sperm counts are declining at all. Studies of sperm quality, especially sperm count, are affected by a number of variables, including measurement methods, temperature, time of day, seasonal variability, and geographic variability (Safe, 2000). For example, as reviewed by Safe (2000), sperm counts among New Yorkers are considerably higher than sperm counts in Californians. Thus, if an initial sample population is disproportionately higher in New Yorkers versus Californians than a subsequent sample population, a true difference in sperm counts may be obscured due to this geographic variability. Safe concluded that present data suggest that it is unknown if sperm counts are going up or down. Thus, it is not yet possible from an epidemiological standpoint to determine if endocrine disruptors are decreasing sperm counts or quality.

In addition to concerns over the validity of the assertion that sperm counts are declining globally, there is also disagreement regarding the impact of specific endocrine disruptors on reproductive health. For example, data showing adverse effects of BPA on sperm count (vom Saal et al., 1998) are countered by better-designed, more comprehensive studies showing no effects of BPA (Ashby et al., 2003). Similar controversies also exist for BPA and prostate health, BPA and the

advancement of puberty, and BPA and aneupoloidy or other genetic disorders (NTP-CERHR Expert Panel Report, 2007). The situation for phthalates essentially parallels that of BPA, such that there is some supportive data for adverse effects in animals, but there is no evidence of a potential impact on humans (Hauser and Calafat, 2005). Thus, despite the outspoken advocacy of the endocrine disruptor hypothesis by certain environmental and health groups, there is no linkage of the limited animal data with actual impacts on human health. If a strong link between "endocrine disruptors" and human health is, in fact, present, it is surprising that such a link has not been clearly delineated in the approximate 50-year history of the use of these compounds.

A second point pertains to potential presence of endocrine disruptors in either breast milk or infant formula and the possibility of harm to infants. While there has been considerable research focused on the measurement of BPA in infant foods, a generally neglected fact is that human milk naturally contains various estrogenic compounds other than BPA. For example, McGarrigle and Lachelin (1983) measured the estrone, estradiol, and estriol content of human milk (both conjugated and unconjugated). The average total concentration of the 3 conjugated hormones from 21 milk samples was 2.021 ppb. The average total concentration of the 3 unconjugated hormones was 0.162 ppb (the concentration of unconjugated estradiol alone was 0.039 ppb). In an attempt to estimate a "BPA-equivalency" of these levels of estrogenic compounds, it is assumed that only the unconjugated form of the hormones is metabolically active. In addition, a review of the various studies of the estrogenic potency of BPA in different model systems (Section 2.2.2 and Table 52 of the NTP-CERHR Expert Panel report), suggests that estradiol is, on average, 10,000 times as potent as BPA, although it is conceded that there is high variability around that average. If the concentration of unconjugated estradiol in human milk is multiplied by a factor of 10,000, then the BPA-equivalent concentration becomes 390 ppb. This is far higher than the trace amounts of BPA (0-20 ppb, on an as-fed basis) that might be detected in infant formula (Table 5 of the NTP-CERHR Expert Panel Report) and does not even include the potential hormonal activities from unconjugated estrone and estriol. If there is no concern for harm regarding the much higher levels of estrogenic activity that already exist naturally in breast milk, then it seems somewhat illogical to worry about the trace amount of estrogenic activity from BPA that may exist in infant formula.

A third point is that rodents and humans have markedly different sensitivities to estrogen. Witorsch (2002) reviewed this issue. In this review, it was noted that estrogen levels attained during late pregnancy in humans are higher by a 100-fold or more than those attained in the mouse. Witorsch further states "modest (2-3-fold) elevations in plasma estradiol in rats and mice are associated with fetal loss, suggesting that these embryos would not survive to term if they were exposed to that seen by human embryos."

A major concern is that many advocates of the "low-dose" BPA hypothesis continue to conduct study after study within the same rodent models. For this reason, very little information exists on BPA's effects on animals that are more closely representative of humans. It is incumbent on researchers to select animal models that are as closely representative of humans as possible. However, because of the increased convenience, relatively lower cost, and greater familiarity of most researchers with rodents compared with other animals, it is possible that these factors, and not scientific validity of the model, are driving the choice of research animals used. The limitations of any animal model as a representative for humans must always be acknowledged in toxicological research. This is especially true for the adverse effects that have been associated with endocrine disruptors, which are often more subtle than other toxicological endpoints that can be readily observed (e.g., tumors). It must be considered that certain rodents may not be

particularly representative of humans with regard to some of the effects that might be associated with endocrine disruptors. As such, extreme caution must be applied in the interpretation of such animal studies.

5. **Confusion regarding the scale of levels of concern employed by the NTP**

The 5-point scale of levels of concern (negligible concern, minimal concern, some concern, concern, and serious concern) makes little sense to the public and is confusing. For example, a rating of "some concern" indicates to the public that governmental concern is present and they will not make the distinctions between the levels of severity of concern intended by NTP (i.e., some concern is not serious concern). Individuals in the general public have to make decisions regarding whether they will buy polycarbonate baby bottles or not and whether they will use a particular food or not. Thus, the only levels of concern that are of interest to consumers are "no concern" or "concern". Consumers will be comfortable with a rating of "no concern" from the government and continue on as usual. If there is "concern" from the government, consumers will expect certain regulatory actions to facilitate consumer protection. They will also stop buying polycarbonate materials and foods that might contain BPA. The middle of the road rating of "some concern" is of little value to consumers, manufacturers, or regulatory agencies. Does "some concern" mean that regulatory action is necessary to protect consumers? If so, NTP should express "concern". If the level of concern is not high enough to necessitate actions to protect the public health, then the NTP must express "no concern." A conclusion of "no concern" can always be modified in the future, as more current scientific information comes in to resolve uncertainties. However, a definitive stand must be taken, one way or the other, regardless of the uncertainties that presently exist in the database. The NTP-CERHR Expert Panel Report and the NTP Draft Brief together represent nearly 500 pages of information but, in the end, give consumers, manufacturers, and regulatory agencies virtually no guidance on what to do about BPA.

Conclusion:

In conclusion, a closer look at the evidence cited by CERHR and NTP suggests that there is little reason to conclude there is "some evidence" of neural and behavioral effects of low dose BPA for pregnant women, infants and children. There is virtually no effect that has been repeated, and many for which contradictory outcomes have been observed. This calls into question whether CERHR's basis of 'more consistent' positive findings for neural and behavioral outcomes is real. Also, the NTP conclusion that there are comparable exposures in the animal studies and human exposures is heavily dependent on one study (contradicted by a second study) that suggests similar outcomes after oral and subcutaneous exposures, and there is ample theory and experience to indicate route of administration is key (as noted by CERHR).

IFC is aware that FDA has tested levels of BPA in infant formula and detected BPA at a range of 0.1 to 13 parts per billion (ppb) (Biles 1997). These levels are considered safe by the FDA and are well below the safety levels set forth by the European Food Safety Authority (EFSA).

The IFC appreciates the opportunity to comment to NTP on this issue. If you have any questions or require clarification of any aspects of these comments, please contact me.

Sincerely,

Mardi K. Mountford, MPH
Executive Vice President

References

Allegaert K, Rayyan M, de Hoon J Tibboel D, Verbesselt R, Naulaers G, Van den Anker JN, Devlieger H. (2006) Contribution of glucuronidation to tramadol disposition in early neonatal life. Bas Clin Pharm Toxicol 98:110-112.

Ashby J, Tinwell H, Haseman J. (1999) Lack of effects for low dose levels of bisphenol A and diethylstilbestrol on the prostate gland of CF1 mice exposed in utero. Reg Toxicol Pharmacol 30:156-166.

Ashby J, Tinwell H, Lefevre PA, Joiner R, Haseman J. (2003) The effect on sperm production in adult Sprague-Dawley rats exposed by gavage to bisphenol A between postnatal days 91-97. Toxicol Sci 74:129-138.

Biles, J. E., McNeal, T. P., Begley, T. H. (1997) Determination of Bisphenol A Migrating from Epoxy Can Coatings to Infant Formula Liquid Concentrates. J. Agric. Food Chem. 45: 4697-4700.

Cagen SZ, Waechter JM, Dimond SS, Breslin WJ, Butala JH, Jekat FW, Joiner RL, Shiotsuka RN, Veenstra GE, Harris LR. (1999) Normal reproductive organ development in CF-1 mice following prenatal exposure to bisphenol A. Toxicol Sci 50:36-44.

Carlsen E, Giwercman A, Keiding N, Skakkebaek NE. (1992) Evidence for the decreasing quality of semen during the past 50 years. Br Med J 305:609-612.

Ceccarelli I, Della Seta D, Fiorenzani P, Farabollini F, Aloisi AM (2007) *Neurotoxicol Teratol.* Estrogenic chemicals at puberty change ERalpha in the hypothalamus of male and female rats. 29(1): 108-115.

Dessi-Fulgheri F, Porrini S, Farabollini F. (2002) Effects of perinatal exposure to bisphenol A on play behavior of female and male juvenile rats. Environ Health Perspect 110 (Suppl 3):403-407.

Ema M, Fujii S, Furukawa M, Kiguchi M, Ikka T, Harazano A. (2001) Rat two-generation reproductive toxicity study of bisphenol A. Reprod Toxicol 15:505-523.

Farabollini F, Porrini S, Della Seta D, Bianchi F, Dessi-Fulgheri F. (2002) Effects of perinatal exposure to bisphenol A on sociosexual behavior of female and male rats. Environ Health Perspect 110 (Suppl 3):409-414.

Funabashi T, Kawaguchi M, Furuta M, Fukushima A, Kimura F (2004) *Psychoneuroendocrinology.* Exposure to bisphenol A during gestation and lactation causes loss of sex difference in corticotropin-releasing hormone-immunoreactive neurons in the bed nucleus of the stria terminalis of rats. 29(4): 475-485

Fujimoto T, Kubo K, Aou S. (2006) Prenatal exposure to bisphenol A impairs sexual differentiation of exploratory behavior and increases depression-like behavior in rats. Brain Res 1068:49-55.

Hauser R, Calafat AM. (2005) Phthalates and human health. Occup Environ Med 62:806-818.

Honma S, Suzuki A, Buchanan DL, Katsu Y, Watanabe H, Iguchi T. (2002) Low dose effect of in utero exposure to bisphenol A and diethylstilbestrol on female mouse reproduction. Reprod Toxicol 16:117-122.

Howdyshell KL, Hotchkiss AK, Thayer KA, Vandenbergh JG, vom Saal FS. Exposure to bisphenol A advances puberty. (1999) Nature 401:763-764.

Howdeshell KL, Furr J, Lambright CR, Wilson VS, Ryan BC, Gray LE. (2008) Gestational and lactational exposure to ethinyl estradiol, but not bisphenol A, decreases androgen-dependent reproductive organ weights and epididymal sperm abundance in the male Long Evans Hooded rat. Toxicol Sci 102:371-382.

Kawai K, Nozaki T, Nishikata H, Aou S, Takii M, Kubo C. (2003) Aggressive behavior and serum testosterone concentration during the maturation process of male mice: The effects of fetal exposure to bisphenol A. Environ Health Perspect 111:175-178.

Kwon S, Stedman DB, Elswick BA, Cattley RC, Welsch F (2000) *Toxicol Sci.* Pubertal development and reproductive functions of Crl:CD BR Sprague-Dawley rats exposed to bisphenol A during prenatal and postnatal development. 55(2): 399-406.

Laviola G, Gioiosa L, Adriani W, Palanza P (2005) *Brain Res Bull.* D-amphetamine-related reinforcing effects are reduced in mice exposed prenatally to estrogenic endocrine disruptors. 65(3): 235-240.

McGarrigle HHG, Lachelin GCL. (1983) Oestrone, oestradiol, and oestriol glucosidurates and sulphates in human puerperal plasma and milk. J Steroid Biochem 18:607-611.

Nagao T, Saito Y, Usumi K, Kuwagata M, Imai K (1999) *Reprod Toxicol.* Reproductive function in rats exposed neonatally to bisphenol A and estradiol benzoate. 13(4): 303-311.

Nagao T, Saito Y, Usumi K, Yoshimura S, Ono H. (2002) Low-dose bisphenol A does not affect reproductive organs in estrogen-sensitive C57BL/6N mice exposed at the sexually mature, juvenile, or embryonic stage. Reprod Toxicol 16:123-130.

Nagel SC, vom Saal F, Thayer KA, Dhar MG, Boechler M, Welshons WV. (1997). Relative binding affinity-serum modified access (RBA-SMA) assay predicts the relative in vivo bioactivity of the xenoestrogens bisphenol A and octylphenol. Environ Health Perspect 105:70-76.

National Toxicology Program-Center for the Evaluation of Risks to Human Reproduction. (2007) NTP-CERHR Expert Panel Report on the Reproductive and Developmental Toxicity of Bisphenol A. Available at: http://www.cerhr.niehs.nih.gov/news/index.html. Accessed: May 28, 2008.

National Toxicology Program. (2008) NTP Draft Brief on Bisphenol A. Available at: http://www. cerhr.niehs.nih.gov/chemicals/bisphenol/BPADraftBriefVF_04_14_08.pdf. Accessed: May 28, 2008.

Negishi T, Kawasaki K, Takatori A, Ishii Y, Kyuwa S, Kuroda Y, Yoshikawa Y (2003) *Environmental Toxicology and Pharmacology.* Effects of perinatal exposure to bisphenol A on the behavior of offspring in F344 rats. 14(3): 99-108.

Negishi T, Kawasaki K, Suzaki S, Maeda H, Ishii Y, Kyuwa S, Kuroda Y, Yoshikawa Y (2004) *Environ Health Perspect.* Behavioral alterations in response to fear-provoking stimuli and tranylcypromine induced by perinatal exposure to bisphenol A and nonylphenol in male rats. 112(11): 1159-1164.

Nishizawa H, Manabe N, Morita M, Sugimoto M, Imanishi S, Miyamoto H (2003) *J Reprod Dev.* Effects of in utero exposure to bisphenol A on expression of RARalpha and RXRalpha mRNAs in murine embryos. 49(6): 539-545.

Nishizawa H, Morita M, Sugimoto M, Imanishi S, Manabe N (2005) *J Reprod Dev.* Effects of in utero exposure to bisphenol A on mRNA expression of arylhydrocarbon and retinoid receptors in murine embryos. 51(3): 315-324.

Pacifici GM, Sawe J, Kager L, Rane A. (1982) Morphine glucuronidation in human fetal and adult liver. Eur J Clin Pharmacol 22:553-558.

Palanza P, Howdeshell K, Parmigiani S, vom Saal FS. (2002) Exposure to a low dose of bisphenol A during fetal life or in adulthood alters maternal behavior in mice. Environ Health Perspect 110(Suppl 3):415-422.

Quinn R. (2005) Comparing rat's to human age: How old is my rat in people years? Nutrition 21:775-777.

Rubin BS, Lenkowski JR, Schaeberle CM, Vandenberg LN, Ronsheim PM, Soto M. (2006) Evidence of altered brain sexual differentiation in mice exposed perinatally to low, environmentally relevant levels of bisphenol A. Endocrinology 147:3681-3691.

Ryan BC, Vandenbergh JG. (2006) Developmental exposure to environmental estrogens alters anxiety and spatial memory in female mice. Horm Behav 50:85-93

Safe SH. (2000) Endocrine disruptors and human health-is there a problem? An update. Environ Health Perspect 108:487-493.

Sharpe RM, Skakkebaek NF. (1993) Are oestrogens involved in falling sperm counts and disorders of the male reproductive tract? Lancet 341: 1392-1395.

Taylor JA, Welshons WV, vom Saal FS. (2008) No effect of route of exposure (oral; subcutaneous injection) on plasma bisphenol A throughout 24 hr after administration in neonatal female mice. Reprod Toxicol 25:169-176.

Tyl Rw, Myers CB, Marr MC, Sloan CS, Castillo NP, Veselica MM, Seely SS, Van Miller JP, Shiotsuka RN, Beyer D, Hentges SG, Waechter JM. (2008) Two-generation reproductive toxicity study of dietary bisphenol A (BPA) in CD-1 (Swiss) mice. Toxicol Sci (epub ahead of print), May 6.

Vandenberg LN, Maffini MV, Wadia PR, Sonnenschein C, Rubin BS, Soto AM (2007) *Endocrinology.* Exposure to environmentally relevant doses of the xenoestrogen bisphenol-A alters development of the fetal mouse mammary gland. 148116-127..

Volkel W, Colnot T, Csanady GA, Filser JG, Dekant W. (2002) Metabolism and kinetics of bisphenol A in humans at low doses following oral administration. Chem Res Toxicol 15:1281-1287.

vom Saal FS, Cooke PS, Buchanan DL, Palanza P, Thayer KA, Nagel SC, Parmigiani S, Welshons WV. (1998) A physiologically based approach to the study of bisphenol A and other estrogenic chemicals on the size of reproductive organs, daily sperm production, and behavior. Toxicol Ind Health 14:239-260.

Witorsch RJ. (2002) Low-dose in utero effects of xenoestrogens in mice and their relevance to humans: an analytical review of the literature. Food Chem Toxicol 40:905-912.